PATENT

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE
PATENT EXAMINING OPERATION

Applicant(s): Glen H. ERIKSON et al.

Serial No: 09/713,177 Group Art Unit: 1637

Filed: November 15, 2000 Examiner: S. Chunduru

Att. Docket No.: E1047/20048 Confirmation No.: 3217

For: TRIPLEX AND QUADRUPLEX CATALYTIC HYBRIDIZATION

APPEAL BRIEF UNDER 37 CFR § 1.192

Mail Stop – Appeal Brief Patents
Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Sir:

I. Introduction

Further to the Notice of Appeal filed May 18, 2004 in response to the February 23, 2004 Final Rejection in which claims 1-63 were finally rejected under 35 U.S.C. § 103(a), Applicants respectfully request reversal of the Final Rejection and allowance of the claims.

A. Real Party in Interest

The real party in interest for this appeal and the present application is the assignee, Ingeneus Corp.

B. Related Appeals and Interferences

Appeals pending in the following application would directly affect or be directly affected by or have a bearing on the Board's decision in the pending appeal: U.S. Patent Application Serial No. 09/885,731, filed June 20, 2001. There are presently no other pending appeals or interferences, known to appellant, appellant's representatives or the assignee, that would directly affect or be directly affected by or have a bearing on the Board's decision in the pending appeal.

C. Status of Claims

Claims 1-63 are pending, are the subject of this appeal and are set forth in the Appendix.

D. Status of Amendments

No amendments have been filed after the appealed Final Rejection dated February 23, 2004.

II. Summary of Invention

Catalytic hybridization is a method in which a large stoichiometric excess of probe is added to target in the presence of a cleaving agent adapted to specifically cleave duplexed probe (see, e.g., specification at page 4, lines 13-21). The cleaved fragments of the probe then dehybridize to provide detectable probe fragments distinguishable from intact probes, and a recycled target available for hybridization with additional intact probes. *Id.* Thus, the target acts as a sort of catalyst for the cleaving step; hence the name "catalytic hybridization". *Id.*

Prior to the present invention, catalytic hybridization was limited to conventional duplex hybridization, wherein the probe and the target are each single-stranded nucleic acid sequences. The present invention provides a method for assaying triplex and quadruplex nucleic acid complexes employing catalytic hybridization, as well as to the complexes formed in the method (see, e.g., claims 1 and 24). Thus, at least one of the probe and the target is double-stranded in the present invention. *Id.* The three or more strands in the complex are bonded solely through Watson-Crick base triplets. *Id.* Such triplets are defined in the specification at page 12, lines 20-22, as "A-T-A, T-A-T, U-A-T, T-A-U, A-U-A, U-A-U, G-C-G and/or C-G-C (including C+-G-C, and/or any other ionized species of base)". Such triplets are not present in traditional

Watson-Crick complexes, regardless of the number of strands in the complex.

III. Issues

Whether claims 1-63 are not obvious under 35 U.S.C. § 103(a) over U.S. Patent No. 5,451,502 (George) in view of J. Mol. Graphics, Vol. 7, pages 218-232, 1989 (McGavin).

IV. Grouping of Claims

Each claim of this patent application is separately patentable, and upon issuance of a patent is to be entitled to a separate presumption of validity under 35 U.S.C. §282. However, pursuant to 37 C.F.R. §1.192(c)(5), for purposes of this appeal, the rejected claims are grouped together in a single group.

V. Argument

Claims 1-63 are not obvious under 35 U.S.C. § 103(a) over U.S. Patent No. 5,541,502 (George) in view of J. Mol. Graph., Vol. 7, pp. 218-232, 1989 (McGavin).

A. No Reasonable Motivation to Modify George with McGavin

George teaches catalytic hybridization of single-stranded probes to single-stranded targets. See George at column 6, lines 1-4 ("The first oligonucleotide used in the present invention is a single-stranded oligonucleotide and has a structure complementary to the nucleic acid sequence being detected.") and at column 9, lines 54-55 ("The assay is initiated by denaturing the sample target molecule to form a single-stranded molecule.").

McGavin was cited by the Examiner in an effort to remedy George's acknowledged failure to "specifically [teach] that the multiplex structure (probe-target complex) is [bonded] solely through Watson-Crick base triplets." See Final Rejection at page 5, first paragraph.

As suggested by the title of McGavin, "A computer graphics study of multistranded DNA models," McGavin discloses theoretical, computer-based models for multiplex nucleic acid sequences, but does not disclose or suggest how the virtual structures depicted by the computer models could be prepared using real nucleic acids. The application of McGavin to reject the claims is based on an improper "obvious-to-try" standard of obviousness. One skilled in the art at the time of the invention would have lacked motivation to employ the purely theoretical teachings of McGavin to modify the primary reference, George, and reach the claimed invention with a reasonable expectation of success. McGavin's virtual teachings provide no guidance regarding how George could be modified to reach the reality of the claimed invention. McGavin reveals nothing regarding hybridization conditions, such as temperature, time, hybridization medium, hybridization promoters, etc. McGavin simply "does not contain a sufficient teaching of how to obtain the desired result." See *Gillette Co. v. S.C. Johnson & Son, Inc.*, 919 F.2d 720, 726, 16 USPQ2d 1923, 1929 (Fed. Cir. 1990), in which the court held:

> [A]n "obvious-to-try" situation exists when a general disclosure may pique the scientist's curiosity, such that further investigation might be done as a result of the disclosure, but the disclosure itself does not contain a sufficient teaching of how to obtain the desired result, or that the claimed result would be obtained if certain directions were pursued. *In re Eli Lilly & Co.*, 902 F.2d 943, 945, 14 USPQ2d 1741, 1743 (Fed. Cir. 1990). However, we have consistently held that "obvious to try" is not to be equated with obviousness under 35 U.S.C. §103.

The Examiner alleges that "[a]n ordinary artisan would have [been] motivated to have added the structural stability of Watson-Crick base pairing of nucleic acid strands in a multiplex structure to the method of George, Jr., because McGavin et al. taught Watson-Crick kind of base pairing as a strong specific interaction between complementary strands and its growing

significance in genetic combination or specificity of interaction between stands." See Final Rejection at page 7, lines 5-14. The Examiner has not identified precisely where McGavin supports this assertion. Presumably, the assertion is based on the following two passages from McGavin:

(1) "Watson-Crick base pairing is clearly a more strongly specific interaction than the additional interaction that we have used to form base tetrads from Watson-Crick base pairs." McGavin at page 230.

(2) "One such model. . . . has proved to be of considerable interest, perhaps particularly in relation to models for genetic recombination." McGavin at page 225.

It should be apparent from the two passages that the motivation alleged by the Examiner is taken out of context. The first passage discloses that the quadruplex (i.e., "tetrad") binding interactions postulated by McGavin are inferior to conventional Watson-Crick duplex binding, and the second passage merely suggests that McGavin's new model might be used as a model to study genetic recombination (presumably mitosis and the like, wherein more than two strands interact on some basis).

The foregoing disclosures of McGavin would not have motivated a person of ordinary skill in the art to modify the teachings of George to reach the claimed invention. Regardless of whether McGavin would have motivated an ordinarily skilled artisan to select a Watson-Crick quadruplex over another type of quadruplex, the Examiner has made absolutely no showing as to why such an artisan would have been motivated to replace the conventional Watson-Crick duplex probe-target complexes of George with the speculative Watson-Crick quadruplexes of

McGavin, which McGavin teaches are based on binding that is not as "strongly specific" as Watson-Crick pairing of two strands. There is no reasonable basis to contend that one skilled in the art would have understood from McGavin that Watson-Crick quadruplexes were more specific and of greater scientific potential than conventional Watson-Crick duplexes.

Moreover, the speculative nature of McGavin, as well as the complete lack of any teaching regarding how to prepare real counterparts of the computer models, would have discouraged a person of ordinary skill in the art from attempting to substitute McGavin's quadruplexes for George's duplexes, such that he or she would have had no reasonable expectation of success. A reference, such as McGavin, containing a statement such as "[w]e have indeed come to think of the use of this core as a model building 'game,'" would not have motivated an ordinarily skilled artisan to modify with a reasonable expectation of success an experimentally proven technology employing a Nobel prize winning duplex model.

Accordingly, the claimed invention is not *prima facie* obvious over George in view of McGavin, as the Examiner has not shown that one of ordinary skill in the art would have been motivated to modify the teachings of George with the teachings of McGavin, and reach the claimed invention with a reasonable expectation of success. See, e.g., MPEP § 2143.

B. McGavin is Improperly Cited Non-enabling Art

A reference relied upon to support an obviousness rejection must provide an enabling disclosure by placing the claimed invention in the possession of the public. See, e.g., *In re Payne*, 606 F.2d 303, 203 USPQ 245, 255 (CCPA 1979). "[I]f the prior art of record fails to disclose or render obvious a method for making a claimed compound, at the time the invention

was made, it may not be legally concluded that the compound itself is in the possession of the public." *In re Hoeksema*, 399 F.2d 269, 274 (CCPA 1968).

In *Hoeksema*, the court held that the examiner had made a *prima facie* case of obviousness by citing a reference disclosing a compound analogous to the claimed compound and a method for making the analogous compound. Applicants in *Hoeksema* submitted an affidavit showing that the claimed compound could not be made by the process disclosed in the applied reference. The court held that the affidavit overcame the *prima facie* showing by showing that the applied reference was not enabling.

In contrast to the applied reference of *Hoeksema*, McGavin does not disclose any process for preparing any chemical compound. As noted above, McGavin discloses theoretical, computer-based models for multiplex nucleic acid sequences based on Watson-Crick bonding, but does not disclose or suggest how to prepare real complexes of nucleic acids corresponding to the virtual models.

Moreover, identifying the chemical formula of a compound that fits within conventional rules of chemistry, as in *Hoeksema*, is far different from building virtual models of multiplex nucleic acids that contravene conventional rules of nucleic acid assembly at the time of the invention, as in McGavin. It is unreasonable to presume that a method for preparing the latter exists from a wholly theoretical reference.

In the Final Rejection at page 6, the Examiner cites MPEP § 2121.04 as supporting an obviousness rejection based on pictures. However, even the cited passage of the MPEP cautions that "the picture must show all the claimed structural features ***and how they are put together***."

[Emphasis added.] While Applicants agree that pictures of a simple mechanical apparatus can inherently reveal how to assemble the apparatus, computer generated pictures depicting a virtual model of nucleic acid strands associated in an unprecedented fashion do not reveal anything about how to prompt nucleic acid strands to assemble in such a fashion. McGavin speculates as to how the nucleobases of multiplex structures might fit together, like pieces of a jigsaw puzzle. Unlike a jigsaw puzzle, however, real world assembly of the multiplex "puzzle" is not simply a matter of snapping adjacent pieces of the puzzle into place.

Thus, the PTO has not sustained its initial burden of making a *prima facie* showing that the claimed compounds and methods for making them were known or obvious in the art at the time of the invention.

C. Consistency of Positions

The Examiner asserts that Applicants' arguments are inconsistent with their previous citation of McGavin in response to the previously pending non-enablement rejection of the claims. Applicants respectfully disagree. A brief review of the prosecution history related to this issue will be helpful in understanding why there is no inconsistency in Applicants present and past positions.

On January 15, 2002, the undersigned conducted an interview with Examiner Chunduru and Primary Examiner Fredman. Although the issues of utility and enablement had not at that time been raised in an Office Action, Examiner Fredman suggested that Applicants clarify how the application enables a useful and credible invention. See January 23, 2002 Amendment at page 8, first paragraph. Examiner Fredman explained that he tends to look more closely at the

level of enablement/credible utility with pioneering inventions exhibiting a high level of novelty, such as the present one. *Id.* More specifically, Examiner Fredman questioned whether Applicants' data suggested triplex or quadruplex formation rather than strand invasion.

Applicants therefore attached to the January 23, 2002 Amendment Rule 132 Declarations of Dr. Jasmine Daksis and Dr. Rick Collins (supplemented on January 25, 2002 with Exhibits B and C inadvertently omitted from the original submission of Dr. Collins' Declaration), as evidence that the data (e.g., the working examples discussed in the specification) were consistent with triplex and quadruplex formation and inconsistent with strand invasion.

The Examiners responded with the June 5, 2002 Final Rejection, which rejected the claims under 35 U.S.C. §§ 101 and 112 as lacking credible enablement and utility. According to that Office Action at the paragraph bridging pages 10-11, a "catalytic hybridization composition and method comprising multiplex structure involving Watson-Crick base pairing is in the [same] class as Cold Fusion, speculative without any structural showing." The December 2, 2002 Advisory Action (last paragraph of attachment) called for "X-ray [crystallographic] or space filling model evidence supporting W-C base [pairing] involving more than two strands" to overcome the rejections.

Applicants in their February 3, 2003 Request for Reconsideration After Entry of RCE cited McGavin (and several other related McGavin references) as only one of several factors suggesting the reasonable credibility of the invention. Applicants never said that McGavin provides even the slightest suggestion regarding how to make a real chemical compound corresponding to the computer models disclosed therein. McGavin was simply cited to provide

space filling model evidence supporting the ***reasonable credibility*** of W-C base bonding involving more than two strands, as requested by the Examiners. See February 3, 2003 Request for Reconsideration at page 4, last paragraph, stating that McGavin's "quantitative calculations suggesting the thermodynamic favorability of four-stranded Watson-Crick complexes lend further credence to the existence of such complexes and the invention claimed by Applicants."

McGavin was only one aspect of Applicants' response to the non-enablement and lack of utility rejections. The present invention is enabled by the specification, which includes working examples suggesting the formation of the claimed multiplexes, and McGavin provides additional evidence supporting the reasonable credibility of such multiplexes.

In summary, Examiners Chunduru and Fredman asserted that the multiplex structures of the invention were theoretically impossible, and Applicants cited McGavin as evidence that multiplex structures were theoretically possible. Referring back to the jigsaw puzzle analogy first mentioned above, the Examiners doubted whether the puzzle pieces could fit together. McGavin was cited as evidence that the puzzle pieces could be arranged in a manner consistent with the claimed invention, without ever asserting that assembly of the multiplex "puzzle" is simply a matter of snapping adjacent pieces of the puzzle into place. Applicants disclose and claim means for assembling the real world counterpart to the virtual puzzle, as well as the assembled counterpart itself.

Accordingly, the claimed invention is not obvious under 35 U.S.C. § 103(a) over George in view of McGavin.

VI. Conclusion

The claims on appeal are not obvious under 35 U.S.C. §103(a) over George in view of McGavin. Accordingly, the Honorable Board of Patent Appeals and Interferences is respectfully requested to withdraw the pending rejection and pass this application on to issuance.

The fee of $165.00 for this Brief, as well as any additional charge or credit, is authorized to be charged to the Deposit Account referenced in the accompanying Form PTO/SB/17. Triplicate copies of this Brief are provided.

Respectfully submitted,

CAESAR, RIVISE, BERNSTEIN,
COHEN & POKOTILOW, LTD.

August 18, 2004

By _____

David M. Tener
Registration No. 37,054
Customer No. 03000
(215) 567-2010
Attorneys for Applicants

Please charge or credit our Account No. 03-0075 as necessary to effect entry and/or ensure consideration of this submission.

Appendix: Claims on Appeal

1. A catalytic hybridization composition comprising:

a probe containing at least one probe nucleobase sequence and at least one scissile linkage sequence;

an enzyme adapted to cleave said at least one scissile linkage sequence;

a nucleic acid target containing at least one target nucleobase sequence associated with said nucleobase sequence of said probe by Watson-Crick bonding to form a multiplex structure; and

a hybridization medium containing said probe, said enzyme and said nucleic acid target,

wherein at least one of said probe nucleobase sequence and said target nucleobase sequence is double-stranded and is bonded to the other of the probe nucleobase sequence or the target nucleobase sequence solely through Watson-Crick base triplets.

2. The composition of claim 1, wherein at least a portion of said multiplex structure is isolated, purified, artificial or synthetic triplex.

3. The composition of claim 1, wherein said probe is single stranded and said target is double stranded.

4. The composition of claim 1, wherein at least a portion of said probe is double stranded.

5. The composition of claim 4, wherein at least a portion of one strand of said probe comprises RNA, mRNA, hnRNA, tRNA or cDNA.

6. The composition of claim 1, wherein at least a portion of said multiplex structure is an isolated, purified, artificial or synthetic quadruplex.

7. The composition of claim 6, wherein a major groove of said probe is placed in a major groove of said target.

8. The composition of claim 6, wherein a major groove of said probe is placed in a minor groove of said target.

9. The composition of claim 1, wherein at least a portion of said probe comprises a nucleic acid or a nucleic acid analogue.

10. The composition of claim 1, wherein at least a portion of said probe comprises a nucleic acid analogue containing an uncharged or partially charged backbone.

11. The composition of claim 1, wherein each nucleobase binds to no more than two other nucleobases.

12. The composition of claim 1, wherein said composition is substantially free of self-hybridized strands.

13. The composition of claim 1, wherein said multiplex structure is substantially free of Hoogsteen binding.

14. The composition of claim 1, wherein said multiplex structure is substantially free of G-G quartets.

15. The composition of claim 1, wherein said probe is 5 to 75 nucleobases.

16. The composition of claim 1, wherein said target is genomic DNA.

17. The composition of claim 1, wherein said target includes a haplotype in genomic DNA.

18. The composition of claim 1, wherein said target comprises PCR amplified products.

19. The composition of claim 1, wherein said multiplex structure is free of solid support.

20. The composition of claim 1, wherein said multiplex structure is bound to a solid support.

21. The composition of claim 20, wherein said solid support is not electrically conductive.

22. The composition of claim 20, wherein said solid support is electrically conductive.

23. The composition of claim 1, wherein said at least one probe nucleobase sequence is from 2 to 30 bases long and said target is at least 8 base pairs long.

24. A method for assaying binding, said method comprising:

providing a probe containing at least one probe nucleobase sequence and at least one scissile linkage sequence;

providing an enzyme adapted to cleave said at least one scissile linkage sequence;

providing a target containing at least one target nucleobase sequence;

combining said probe, said enzyme and said target in a hybridization medium further containing water, a buffer and at least one promoter;

incubating said hybridization medium to hybridize said probe nucleobase sequence to said target nucleobase sequence by Watson-Crick bonding to form a multiplex, wherein at least one of said probe nucleobase sequence and said target nucleobase sequence is double-stranded and is bonded to the other of the probe nucleobase sequence or the target nucleobase sequence solely through Watson-Crick base triplets;

cleaving hybridized probes at said at least one scissile linkage to provide unbound probe fragments; and

detecting said unbound probe fragments to assay binding between said probe and said target.

25. The method of claim 24, wherein an incubation temperature is from 2°C to 60°C.

26. The method of claim 24, wherein said hybridization medium is buffered to a pH of about 5 to about 9.

27. The method of claim 24, wherein said at least one promoter is an intercalating agent.

28. The method of claim 27, wherein said at least one promoter is an intercalating fluorophore, and a fluorescent intensity of a test medium containing said multiplex structure is directly correlated with a binding affinity of said probe for said target.

29. The method of claim 28, wherein said intercalating fluorophore is a member selected from the group consisting of YOYO-1, TOTO-1, ethidium bromide, ethidium homodimer-1, ethidium homodimer-2 and acridine.

30. The method of claim 24, wherein said at least one promoter is tethered to said probe.

31. The method of claim 24, wherein said at least one promoter is a monovalent cation.

32. The method of claim 24, wherein said at least one promoter is a cation having a valency greater than one.

33. The method of claim 32, wherein said at least one promoter is at least one member selected from the group consisting of alkali metal cations, alkaline earth metal cations, transition metal cations, $Co(NH_3)_6^{+3}$, trivalent spermidine and tetravalent spermidine.

34. The method of claim 32, wherein said cation is K^+ or Na^+ provided at a concentration of 40 mM to 200 mM.

35. The method of claim 24, wherein said target is provided in said hybridization medium before said probe, and wherein said probe is provided in dehydrated form prior to rehydration by contact with said hybridization medium.

36. The method of claim 24, wherein said incubation time is not more than about 24 hours.

37. The method of claim 24, wherein probe-target hybridization is detected as a change in a fluorescent, chemiluminescent, electrochemiluminescent or electrical signal.

38. The method of claim 37, wherein an intensity of said signal is correlated with a binding affinity between said probe and said target.

39. The method of claim 38, wherein said probe is covalently labeled with a non-intercalating fluorophore and said intensity is inversely correlated with said binding affinity.

40. The method of claim 39, wherein said non-intercalating fluorophore is a member selected from the group consisting of biotin, rhodamine and fluorescein.

41. The method of claim 37, wherein said method is a homogeneous assay.

42. The method of claim 24, wherein said probe is covalently labeled with a marking agent on a first side of said at least one scissile linkage and a quenching agent on a second side of said at least one scissile linkage, wherein said quenching agent quenches a signal of said marking agent when said probe is intact and does not quench said signal after said probe is cleaved.

43. The method of claim 24, wherein said probe is covalently labeled with a marking agent on a first side of said at least one scissile linkage and a amplification agent on a second side of said at least one scissile linkage, wherein said amplification agent amplifies a signal of said marking agent when said probe is intact and does not amplify said signal after said probe is cleaved.

44. The method of claim 24, further comprising separating intact probes from said probe fragments.

45. The method of claim 24, wherein a ratio of said probe to said target is from 100:1 to 1000:1.

46. The method of claim 24, wherein concentrations of said probe and said target are not more than 5×10^{-10} M.

47. The method of claim 24, wherein said at least one promoter is a minor groove nucleic acid binding molecule, which binds in a non-intercalating manner and binds with an association constant of at least 10^3 M^{-1}.

48. The method of claim 24, wherein conditions of hybridization are subject to transitory or periodic changes.

49. The method of claim 48, wherein the changes are caused by applying a force.

50. The method of claim 49, wherein the force applied is electric, magnetic or mechanical.

51. The method of claim 24, wherein cleaved probe and unhybridized probe remain in solution.

52. The method of claim 24, wherein said enzyme will cleave only RNA sequences of nucleotides in a multiplex structure.

53. The method of claim 24, wherein said enzyme will cleave only nucleobases having predetermined backbone characteristics.

54. The method of claim 24, wherein a backbone structure of said probe is composed entirely of RNA.

55. The method of claim 54, wherein said at least one scissile linkage sequence is about 2 to about 12 nucleotides in length.

56. The method of claim 24, wherein said probe contains at least one interspersed sequence that is not cleavable by said enzyme.

57. The method of claim 56, wherein said at least one interspersed sequence comprises DNA or DNA analogues.

58. The method of claim 57, wherein said at least one interspersed sequence comprises nucleotide residues selected from the group consisting of phosphonates, phosphotriesters, phosphoroamidates and 2'-0 alkyl and aryl ribonucleotide.

59. The method of claim 24, further comprising suppressing non-specific cleavage of the probe with at least one single-stranded ribonuclease inhibitor selected from the group consisting of vanadate, RNAsin, and Inhibit - ACE.

60. The method of claim 24, wherein said enzyme is RNAaseH.

61. The method of claim 60, wherein said RNAaseH enzyme is obtained from E. coli.

62. The composition of claim 1, wherein said probe comprises an electrically, electromechanically or optically active reporter group adapted to emit a detectable signal.

63. An electrical circuit comprising the composition of claim 1.